US Case G-33347/P1/9942

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   on 11th September 2003

   under the number A 1433/2003

   and the official certificate attached thereto.

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Dated this 22nd day of February 2006

J. Crook

AUSTRIAN PATENT OFFICE

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The Austrian Patent Office herewith confirms that

the company **SANDOZ G.m.b.H.**
in A-6250 Kundl , Biochemiestrasse 10 (Tyrol)

filed a patent application relating to

"Organic compounds"

on **11th September 2003**

and that the attached description is completely identical to the original description filed simultaneously with this patent application.

Austrian Patent Office
Vienna, 16th July 2004
The President
per pro
(signature)

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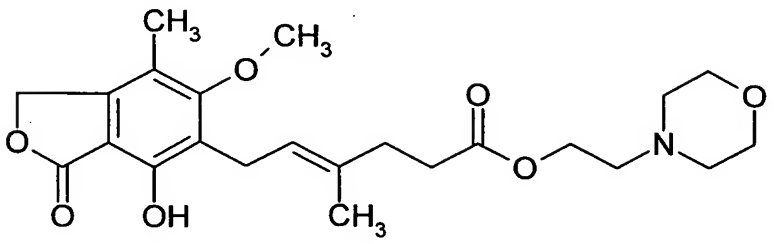
Organic compounds

The present invention relates to a new process for the production of mycophenolate mofetil of formula



As a result of its immunosuppressive, antiinflammatory, antiviral and anti-tumour activity, mycophenolate mofetil [mycophenolic acid 2-(4-morpholinyl)ethyl ester, or (4E)-6-(1,3-dihydro-4-hydroxy-6-methoxy-7-methyl-3-oxo-5-isobenzofuranyl)-4-methyl-4-hexenoic acid-2-(4-morpholinyl)-ethyl ester according to Merck Index / 13th Edition / Monograph number 6352] is used as the active component of pharmaceutical preparations, e.g. in the prevention of rejection reactions following transplantations, in the treatment of autoimmune diseases, psoriasis, inflammatory disorders such as rheumatic arthritis, as well as viral illnesses and tumours.

The production of mycophenolate mofetil from the fermentation product of mycophenolic acid and 4-(2-hydroxyethyl)morpholine is a demanding procedure because of the basic function of the morpholine moiety and the polyfunctionality of the mycophenolic acid.

For example, a process is described in EP 0649422 for esterification without a catalyst with relatively long reaction times, which do not appear to be attractive from an economical point of view.

US 4753935 describes the production of mycophenolate mofetil via an acid chloride of mycophenolic acid which is produced with thionyl chloride, or via activation with carbodiimide. According to EP 0649422, these two methods are regarded as having little suitability or none at all for producing mycophenolate mofetil in pharmaceutically acceptable purity, especially because of impurities that arise.

It has now surprisingly been found that mycophenolate mofetil can be produced in high purity and in a high yield by modifying, in accordance with the invention, the above-described acid chloride process. This new process, as is described in the present invention, is thereby extremely attractive in consideration of economic yield.

One aspect of the invention therefore comprises a process for the production of mycophenolate mofetil, whereby a reactive derivative of mycophenolic acid is prepared in an inert solvent and is reacted with 4-(2-hydroxyethyl)morpholine, and the resulting mycophenolate mofetil is isolated from the reaction mixture, characterised in that

I) 4-(2-hydroxyethyl)morpholine is added under controlled conditions to the solution of the reactive derivative of mycophenolic acid, whereby the reaction takes place under acidic reaction conditions, and

II) isolation of mycophenolate mofetil is effected by forming an acid addition salt and subsequently releasing the free base.

In a further aspect of the invention, the process for the production of mycophenolate mofetil includes the following steps:

a) activation of mycophenolic acid by forming a reactive derivative by known methods,

b) reacting the reactive derivative of mycophenolic acid with 4-(2-hydroxyethyl)morpholine by esterifying to mycophenolate mofetil under acidic reaction conditions,

c) isolating mycophenolate mofetil through the formation of an acid addition salt, and

d) releasing the free base of mycophenolate mofetil from the acid addition salt by known methods.

Using the process according to the invention, mycophenolate mofetil is obtained as the free base in a very high degree of pharmaceutically acceptable purity.

A further aspect of the invention comprises the production of the crystalline addition salt of mycophenolate mofetil with oxalic acid and its use as an intermediate in the production of mycophenolate mofetil as the free base or as the pharmaceutically acceptable salts thereof with pharmaceutically acceptable purity.

By an "inert solvent" as described in the present invention is understood a solvent which is inert to the reaction partners under the reaction conditions described here, especially a solvent that is immiscible or only very poorly miscible with water, such as acetic acid (C_1-C_4) alkylester, for example ethyl acetate or halogenated hydrocarbons, for example dichloromethane, optionally in the presence of a cosolvent. Suitable cosolvents are amides,

such as N,N-dimethylformamide or N,N-dimethylacetamide, or cyclic amides, for example N-methylpyrrolidone, or cyclic ureas, for example 1,3-dimethyl-2-imidazolidinone (DMEU) or 1,3-dimethyl-3,4,5,6-tetrahydro-2(1H)-pyrimidinone (DMPU).
Expecially preferred combinations are mixtures of ethyl acetate and N,N-dimethylformamide, and of dichloromethane and N,N-dimethylformamide.

By "pharmaceutically acceptable salts" of mycophenolate mofetil, as described in the present invention, is understood for example the hydrochloride.

The activation of mycophenolic acid may take place for example by Vilsmeier technology (according to Vilsmeier A., Chem.-Ztg. *[Chem Journal]*75, 133-135, 1951; CD Römpp Chemie Lexikon – Version 1.0, Stuttgart/New York: Georg Thieme Verlag 1995), for example with an isolated Vilsmeier reagent, for example N,N-dimethyl(chloromethylene)iminium chloride, or by *in situ* activation, for example with an organic amide such as N,N-dimethylformamide or N,N-dimethylacetamide in combination with a halogenation agent, for example oxalyl chloride.

The amount of activation reagent can be kept small for economic reasons. For example, 1 to 1.5 molar equivalents, preferably 1.05 to 1.3 molar equivalents, of Vilsmeier reagent, or Vilsmeier reagent prepared *in situ*, are used. The *in situ* preparation of Vilsmeier reagent in the combination N,N-dimethylformamide and oxalyl chloride is preferred, whereby the ratio of N,N-dimethylformamide to oxalyl chloride is 10 : 1 to 1 : 2, preferably 1 : 1 to 1 : 1.2.

Suitable solvents for activation of the mycophenolic acid by the process according to the invention are the above-described inert solvents, optionally in the presence of a cosolvent as listed above.

The activation of mycophenolic acid may be carried out at temperatures of -20°C to room temperature, for example from -20°C to +25°C, for example from -10°C to +10°C, preferably at 0°C.

Alternatively, the activation of mycophenolic acid may also take place with current halogenation agents, for example thionyl chloride, or halogenated phosphorus compounds, for example phosphorus trichloride and phosphorus pentachloride, or 2,4,6-trichloro-1,3,5-triazine or 5-chloro-4,6-dimethoxy-1,3,5-triazine.

The reactive derivative obtained through activation of mycophenolic acid is an activated carboxylic acid derivative, for example an acid halide, preferably an acid chloride.

The subsequent reaction of the reactive derivative of mycophenolic acid, for example the acid chloride, with 4-(2-hydroxyethyl)morpholine is carried out in such manner that the alcohol is added to the solution of the acid chloride under controlled conditions, for example over ca. 2 to ca. 180 minutes, preferably over ca. 10 to ca. 60 minutes, so that the excess of free alcohol is kept at a relatively low level.
The hydrochloric acid being released in the acylation reaction thereby lowers the basicity of the reaction mixture, so that under the resultant acidic reaction conditions the formation of by-products is greatly suppressed. The reaction progresses more or less quantitatively with little excess of 4-(2-hydroxyethyl)-morpholine.

For the esterification reaction described here, for example 0.8 to 2.5 molar equivalents, preferably 1.05 to 1.3 molar equivalents, of 4-(2-hydroxyethyl)-morpholine are added.

According to a preferred variant of the process according to the invention, the esterification reaction is started at lower temperatures and is carried out at higher temperatures in order to complete the reaction.

In a further preferred variant, 4-(2-hydroxyethyl)morpholine is added at temperatures of 0°C to 40°C, for example from 10°C to 30°C, preferably at temperatures around 20°C, and the reaction is brought to an end at temperatures of 40°C to 120°C, preferably at 40°C to 60°C.

The reaction times for the esterification reaction according to the invention are, for example, ca. 3 to ca. 20 hours, preferably ca. 5 to ca. 15 hours.

A further aspect of the invention is the subsequent isolation of mycophenolate mofetil from the acidic reaction mixture by forming an acid addition salt, for example the oxalate or hydrochloride, and subsequently releasing the free base.

If the above-described esterification reaction is carried out in a corresponding apolar solvent, for example an acetic acid (C_1-C_4)-alkylester, for example ethyl acetate, mycophenolate mofetil precipitates from the acidic reaction mixture directly as the hydrochloride.
The mycophenolate mofetil hydrochloride is isolated by known methods, for example by filtration.

Release of the free base may be carried out for example in a two-phase system - water with an organic solvent - with the assistance of a base such as sodium or potassium hydroxide, an

alkali carbonate or alkali hydrogen carbonate, and mycophenolate mofetil is crystallised for example from ethyl acetate or an alcohol such as ethanol or isopropanol or a ketone such as acetone or a combination of said solvents, optionally after evaporation of the solvent.
The preferred organic solvent for release of the free base is ethyl acetate.

In a preferred embodiment of the process according to the invention, the crystalline addition salt of mycophenolate mofetil is formed with oxalic acid, and is used as an intermediate in the production of mycophenolate mofetil as the free base.

The oxalate of mycophenolate mofetil is notable for its poor solubility in current organic solvents. It is therefore especially suitable for isolating mycophenolate mofetil from solvents in which both the free base and the hydrochloride of mycophenolate mofetil are very soluble.

For example, the oxalate of mycophenolate mofetil can be isolated from dichloromethane optionally in combination with N,N-dimethylformamide. If necessary, a further solvent, for example a (C_1-C_4)-alcohol, for example methanol, or a ketone, for example acetone, is added.

In a preferred embodiment of the process according to the invention, therefore, isolation of mycophenolate mofetil can also be carried out directly from acylation solutions, as are produced preferably by using dichloromethane.
After the esterification reaction has taken place as described above, the acidic reaction mixture is neutralised, i.e. brought to a pH value of 5 - 9, preferably 6 - 8 with a base, for example sodium or potassium hydroxide, or an alkali carbonate or alkali hydrogen carbonate, and the oxalate of mycophenolate mofetil is crystallised by adding oxalic acid, for example a solution of oxalic acid, for example a 10 to 30% solution in acetone or in an alcohol such as methanol.

In another preferred embodiment, mycophenolate mofetil can be isolated from a reaction mixture as it is obtained for example in ethyl acetate, after neutralisation, and obtained as the oxalate by adding oxalic acid, as described above.

The oxalate of mycophenolate mofetil, as it is produced by the process according to the invention, is present in crystalline form with high purity, i.e. with HPLC purity of for example more than 98%, preferably more than 99%.
If required, the crystalline oxalate of mycophenolate mofetil may also exist as the hydrate or solvate.

The free base is released from the oxalate by the method described above.

By employing the isolation of mycophenolate mofetil from the acidic reaction mixture, as described in the process according to the invention, through the formation of the acid addition salt, for example the oxalate or hydrochloride, preferably the oxalate, and the subsequent release of the free base, mycophenolate mofetil is obtained in a high yield and in very pure form, i.e. with a HPLC purity of more than 99%, preferably more than 99.5%, and with a content of by-products of no more than 0.1%, preferably no more than 0.05%.

An additional advantage of the process according to the invention is the relatively short reaction times during the esterification reaction.

The process according to the invention is therefore economically attractive when producing mycophenolate mofetil as a free base or as the pharmaceutically acceptable salts thereof, for example the hydrochloride, in pharmaceutically acceptable pure form.

The following examples are intended to illustrate the invention more fully, without limiting its scope. All temperatures are given in degrees celsius.

Example 1:

Production of mycophenolate mofetil through the formation of the mycophenolate mofetil hydrochloride

Example 1a:

Production of mycophenolate mofetil hydrochloride

9.094 g of mycophenolic acid are added at room temperature to a mixture of 140 ml of ethyl acetate and 3.12 ml of N,N-dimethylformamide. The suspension is cooled to 0°, and subsequently 3.05 ml of oxalyl chloride, dissolved in 15 ml of ethyl acetate, are added dropwise over ca. 20 minutes, whereby a solution is obtained. The solution is stirred for a further 140 minutes at 0° to 2° and then heated to 20°. Then, a solution of 4.16 ml of 4-(2-hydroxyethyl)morpholine in 20 ml of ethyl acetate is added over the course of ca. 20 minutes at 20° to 22°, whereby a suspension forms already after the first drops. The suspension is heated to 60° and is stirred at this temperature for ca. 20 hours. The title compound is then isolated by filtering the hot suspension through a suction filter, washed twice, each time with 25 ml of ethyl acetate heated to 60°, and dried in a vacuum over night at room temperature.
Weight of mycophenolate mofetil hydrochloride: 11.85 g
HPLC purity: 97.8%

Example 1b:

Production of mycophenolate mofetil as the free base

10 g of the mycophenolate mofetil hydrochloride obtained in example 1a are suspended in 100 ml of ethyl acetate. 50 ml of water are added and stirred until a two-phase solution is obtained. Then, the pH value is adjusted from ca. 2.0 to pH 7.4 to 7.5 using ca. 30 ml of saturated $NaHCO_3$ solution, the phases are separated and the organic phase is washed first of all with a mixture of 50 ml of water and 2.5 ml of saturated $NaHCO_3$ solution, and then twice with 50 ml of water each time. The ethyl acetate phase is then mixed with 1 g of activated carbon and stirred for 10 minutes at room temperature. The activated carbon is removed by filtration, the filter cake washed with 10 ml of ethyl acetate, and the combined phases are concentrated to ca. 40 g by evaporation in a vacuum. The residue is seeded with the title compound, the resulting suspension is stirred for one hour at room temperature and then cooled to ca. -20°. After standing over night in a refrigerator at ca. -20°, the title compound is isolated through a suction filter, washed with 5 ml of cold ethyl acetate and dried over night in a vacuum.

Weight of mycophenolate mofetil as the free base: 6.71 g

HPLC purity: 99.5%

Example 2:

Production of mycophenolate mofetil through the formation of the mycophenolate mofetil oxalate

Example 2a:

Production of mycophenolate mofetil oxalate

9.094 g of mycophenolic acid are dissolved at room temperature in a mixture of 175 ml of dichloromethane and 1.56 ml of N,N-dimethylformamide. The solution is cooled to 0°, and a solution of 3.05 ml of oxalyl chloride in 10 ml of dichloromethane is added dropwise to the thin suspension over the course of ca. 15 minutes, whereby a clear solution is again obtained. After the addition is complete, the mixture is stirred for a further 30 minutes at 0°. The solution is then brought to ca. 20°. A solution of 4.16 ml of 4-(2-hydroxyethyl)morpholine in 20 ml of dichloromethane is added dropwise over the course of ca. 55 minutes. The dropping funnel is rinsed with 5 ml of dichloromethane and the solution is subsequently boiled under reflux for 15 hours. Afterwards, the solution is cooled to 0°, mixed with 1.5 ml of methanol, and after stirring for 30 minutes, is mixed with 85 ml of water. The two-phase solution is

stirred for ca. 5 minutes at 15° to 20° and then the pH value is set at ca. 3.2 with saturated $NaHCO_3$ solution. The mixture is filtered, the phases are separated, 50 ml of water is added to the dichloromethane phase, and the pH value is set at ca. 7.5 with saturated $NaHCO_3$ solution. The phases are separated and the dichloromethane phase is washed with 50 ml of water, whereby the pH value is again adjusted to 7.5 with saturated $NaHCO_3$ solution, and the dichloromethane phase is subsequently washed twice more, each time with 50 ml of water. 2 g of activated carbon are added to the dichloromethane phase, the suspension is stirred for 10 minutes and the carbon is filtered off through a suction filter. The filter cake is washed with 10 ml of dichloromethane and mixed with the filtrate. This mixture consisting of the filtrate and the washed phase is subsequently added dropwise over the course of ca. 30 minutes to a solution of 2.81 g of water-free oxalic acid in 14.1 ml of acetone, whereby the title compound crystallises. The suspension is stirred for one hour at room temperature, cooled with ice for a further 2 hours, then the title compound is isolated through a suction filter, washed twice, each time with 25 ml of dichloromethane, and dried over night at room temperature in a vacuum drying chamber.

Weight of mycophenolate mofetil oxalate: 13.45 g

HPLC purity: 99.4%

Content of oxalic acid (HPLC): 19.8% (percent by weight)

Example 2b:

Production of mycophenolate mofetil as the free base

10 g of the compound from example 2a are suspended in a mixture of 50 ml of water and 100 ml of ethyl acetate. The pH value is then adjusted to 7.4 to 7.5 with 10% $KHCO_3$ solution, whereby a two-phase solution is produced. The mixture is stirred for ca. 5 minutes, and the phases are subsequently separated. The ethyl acetate phase is subsequently washed with a mixture of 50 ml of water and 2 ml of 10% $KHCO_3$ solution, followed by 2 washes each time with 50 ml of water. The ethyl acetate phase is concentrated in a vacuum on a rotary evaporator to 25 g, seeded with mycophenolate mofetil, and the resulting suspension is stirred first of all for 1 hour at room temperature, then for 1 hour with ice cooling, and is subsequently stored over night in a refrigerator at ca. -20°. The crystals are subsequently isolated through a suction filter, washed with 5 ml of cold ethyl acetate and dried in a vacuum drying chamber at room temperature.

Weight of mycophenolate mofetil as the free base: 7.01 g

HPLC purity: 99.7%

Example 3

Characterisation of mycophenolate mofetil oxalate

1 g of the mycophenolate mofetil oxalate obtained from example 2a is dissolved in a mixture of 10 ml of acetone and 3 ml of water at 60°. 30 ml of acetone are added to the solution whilst rotating, and the resulting suspension is left to stand for one hour at room temperature. The mixture is stirred gently for 30 minutes, then left to stand for one hour whilst cooling with ice. The crystals are subsequently isolated through a suction filter and washed in two portions with a total of 4 ml of acetone. Drying is carried out at room temperature in a vacuum.
Content of oxalic acid (HPLC): 17.1% (percent by weight); theory: 17.2% (percent by weight)
Melting point: 164°-165°

Example 4:

Production of mycophenolate mofetil through the formation of the mycophenolate mofetil oxalate

Example 4a:

Production of mycophenolate mofetil oxalate

13.64 g of mycophenolic acid are dissolved at room temperature in a mixture of 196.5 ml of dichloromethane and 3.54 ml of N,N-dimethylformamide. The solution is then cooled to 0°. A solution of 3.92 ml of oxalyl chloride in 15 ml of dichloromethane is added dropwise through a dropping funnel during ca. 20 minutes whilst stirring. The dropping funnel is rinsed with 10 ml of dichloromethane and the solution is stirred for ca. one hour at 0°. The solution is then brought to 20°. Then, a solution of 6.24 ml of 4-(2-hydroxyethyl)morpholine in 30 ml of dichloromethane is added to this solution dropwise over the course of ca. 30 minutes, and afterwards the dropping funnel is rinsed with 10 ml of dichloromethane. The mixture is subsequently boiled under reflux for 12 hours and then cooled to room temperature. The solution is mixed with 127 ml of water, stirred for ca. 30 minutes, and then extracted with 200 ml of saturated $NaHCO_3$ solution. The organic phase is filtered through a K-150 filter sheet (available commercially from Seitz Filter Werke GmbH, Germany). The aqueous phase is extracted with 50 ml of dichloromethane, and the organic phases are combined and extracted with a mixture of 75 ml of water and 10 ml of saturated $NaHCO_3$ solution. The phases are separated and the dichloromethane phase is extracted twice more, each time with 75 ml of water. 3 g of activated carbon are added to the dichloromethane phase, the suspension is stirred for ca. 10 minutes, the activated carbon is filtered off through the K-150 filter sheet, and the activated carbon is then washed with 15 ml of dichloromethane. A solution of 4.22 g of oxalic acid in 15 ml of methanol is added to the combined organic phases, the solution is

seeded with seed crystals of mycophenolate mofetil oxalate, the solution which is becoming thicker is left to stand at room temperature for ca. one hour, and is then stirred for ca. 3 hours whilst cooling with ice. The product is isolated through a suction filter, washed with 76 ml of dichloromethane in two portions, and dried over night in a vacuum at room temperature.

Weight of mycophenolate mofetil oxalate: 18.67 g

HPLC purity: 99.6%

Example 4b:

Production of mycophenolate mofetil as the free base

10 g of mycophenolate mofetil oxalate from example 4a are converted into the mycophenolate mofetil free base as in example 2b.

Weight of mycophenolate mofetil as the free base: 6.99 g

HPLC purity: 99.9%